- exposing the sample to the action of a gradient that, at least partially, denatures the amplified nucleic acid in the sample and that effects variation in the spectroscopically measurable parameter of the probe, creating a measurable signal;

- detecting the measurable signal; and

- optionally carrying out the amplification reaction and the qualitative and quantitative analysis without opening the sealed reaction chamber.

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68. 2 The process according to Claim 67, 1 wherein the spectroscopically measurable parameter of the probe is at least one luminescent or fluorescent dye, and the probe includes a nucleic acid portion, which interacts with the <u>in vitro</u> amplified nucleic acid during the denaturation accompanied by a change in the measurable signal.

69. 3 The process according to Claim 67, 1 wherein the measurable signal is detected (a) using wave length variation, shift in luminescence or fluorescence intensity, variation in fluorescence polarization, variation in excited state lifetime, or a combination thereof, or (b) using the principle of energy transfer, or (c) through a concentration effect.

70. 4 The process according to Claim 67, 1 wherein the spectroscopically measurable parameter includes a plurality of dyes distinguishable from each other spectroscopically.

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71. 5 The process according to Claim 70, 4 wherein a laser excites luminescence of the dyes.

72. 6 The process according to Claim 67, 1 wherein the reaction mixture includes at least one co-amplified nucleic acid standard, the sequence of which is homologous to a sequence to be analyzed, with the exception of at least one point mutation.

73. 7 The process according to Claim 67, 1 that includes at least one co-amplified nucleic acid standard having a primer region, the sequence of which is homologous to the primer region of the amplified nucleic acid.

74. 8 The process according to Claim 73, 7 wherein the nucleic acid standard is a natural component of the amplified nucleic acid.

75. The process according to Claim 67, wherein amplification is carried out (a) in homogenous phase or (b) using a primer attached to a solid phase, the amplified nucleic acid hybridizes with the probe, and the analysis is determined either attached to the solid phase or within the homogenous phase.

76. The process according to Claim 73, wherein the probe is is at least one molecule of fluorescent dye linked to a nucleic acid molecule, the sequence of which is identical or homologous to the amplified nucleic acid to be or to the co-amplified nucleic acid standard.

77. 11 The process according to Claim 75, 10 wherein the fluorescent dye linked to the nucleic acid molecule is added to the

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reaction mixture after completing amplification, and is hybridized with the amplified nucleic acid by thermal denaturation with subsequent renaturation.

~~78.~~ 12 The process according to Claim ~~76~~ 10, wherein the fluorescent dye linked to the nucleic acid molecule is added to the reaction mixture prior to completing amplification, and the probe is a non-amplifiable double-stranded RNA or a non-amplifiable chemically modified nucleic acid.

~~79.~~ 13 The process according to Claim ~~67~~ 1, wherein a primer of a primer pair is used for the amplification, which primer encodes a G:C-rich region at the 5' terminus ~~of preferably from 15 to 20 G:C residues~~.

80. The process according to Claim 67, wherein the probe is an oligo- or polynucleotide having at least two chemical structural elements that can cleave a stable double bond of the oligo- or polynucleotide and, optionally, link the stable double bond to another position on the oligo- or polynucleotide by absorbing electromagnetic radiation, excitation-effected emitting of electromagnetic radiation, or a combination thereof.

~~81.~~ 15 The process according to Claim ~~80~~ 14, wherein the chemical structural elements have a chromophoric system.

~~82.~~ 16 The process according to Claim ~~81~~ 15, wherein the chromophoric system luminesces via a dye substituent thereon.

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83. The process according to Claim 80, wherein the chemical structural elements are photochemical crosslinkers.

84.18 The process according to Claim 83,17 wherein the photochemical crosslinkers are psoralene or a psoralene derivative.

85.19 The process according to Claim 80,14 wherein spacing between the two chemical structural elements is between 8 to 12 nucleotide positions.

86.20 The process according to Claim 67,1 wherein the reaction means comprises a plurality of recesses in a sheet system, each recess thermally weldable, accommodates ready-for-use reagent mixtures in lyophilized or matrix-bound form, and permits direct optical measurement.

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87.21 The process according to Claim 86,20 wherein the reagent mixtures are stored in spatially separated matrices, and, subsequent to sealing the reaction chamber, are introduced into the reaction process.

88.22 The process according to Claim 67,1 wherein the analysis is effected by microtitration.

89.23 The process according to Claim 67,1 wherein the gradient is a time-controlled temperature gradient, and the variation of the spectroscopically measurable parameter is monitored as a function of time, temperature, or time and temperature.

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90. The process according to Claim 89, wherein the analysis is by temperature gel electrophoresis, chromatography, or directly in homogenous solution, or a combination thereof.

91. The process according to Claim 90, wherein the presence, number, homology, or combination thereof of the amplified nucleic acid depends on the monitored spectroscopically measurable parameter.

92. The process according to Claim 67, wherein the analysis is effected using a data processing system.

93. The process of claim 67 wherein the probe is an oligo- or polynucleotide having at least one non-naturally occurring chemical structural element that can cleave a stable double bond of the oligo- or polynucleotide and, optionally, link the stable double bond to another position on the oligo- or polynucleotide by absorbing electromagnetic radiation, excitation-effected emitting of electromagnetic radiation, or a combination thereof, and wherein said structural element is not a purine or pyrimidine substituent of naturally occurring nucleotide components.

94. The process of claim 93 wherein the chemical structural element is psoralene or a psoralene derivative.

95. The process of claim 93 wherein the chemical structural element luminesces.

30 27
96. The process of claim 93 wherein one of the chemical structural elements is located 8 to 12 nucleotides away from another of the chemical structural elements.

31 1
97. The process of claim 67 wherein the reaction means includes (A) at least one multiple-well-containing sheet, each well being a reaction chamber that includes the probe for and lyophilized amplification reagents and (B) a sealing sheet cooperating with the multiple-well-containing sheet in a manner independently sealing each reaction chamber with a seal that becomes an interior surface of the reaction chamber.

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32 31
98. The process of claim 97 wherein the reagents are present in at least one water-soluble matrix.

33 32
99. The process of claim 98 wherein the matrix includes a stabilizer.

34 32
100. The process of claim 98 wherein the matrix includes a sugar.

35 32
101. The process of claim 98 wherein the matrix includes trehalose or saccharose.

36 32
102. The process of claim 98 wherein the reagents include amplification primers, buffer components, at least one polymerase, and co-factors.

37 31
103. The process of claim 97 wherein the reagents include amplification primers, buffer components, at least one polymerase, and co-factors.

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104. 38 The process of claim 97 31 wherein at least one reaction chamber of the well-containing sheet includes a reagent/probe-containing matrix and the chamber interior surface of the corresponding seal includes hybridization reagents.

105. 39 The process of claim 97 31 wherein the reaction means is composed of kit systems.

106. 40 The process of claim 67 1 including computer-controlled, time-dependent thermostating of the reaction chamber.

107. 41 The process of claim 67 1 including optical-excitation-effecting emitting of a fluorescence signal and optical detection of the fluorescence signal.

108. 42 The process of claim 107 41 wherein the excitation is by a laser.--

REMARKS

The present claims are 67-108.

The present claims represent cancelled claims 1-26 and 39-54 written in order to more clearly define the present invention.

Applicants respectfully submit that a new oath or declaration is not required. The oath identifies the application by its PCT number. This is entirely appropriate and permissible under the statute, rules, and regulations.

Applicants submit herewith a new Abstract as required in the outstanding Office action.